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AUTHOR(S):

Kubota, Shin

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Cnidome and Growth of a Medusa of *Cirrholovenia tetranema* (Leptomedusae, Cirrholoveniidae) in Japan

SHIN KUBOTA

Seto Marine Biological Laboratory, Kyoto University,
Shirahama, Wakayama 649-22, Japan

With Text-figures 1-9 and Tables 1-4

Abstract The cnidome, growth of a medusa, as well as the gametes and spawning time, of *Cirrholovenia tetranema* Kramp (Leptomedusae, Cirrholoveniidae) were studied for the first time. The medusae were collected from six localities: Tsushima Island (Nagasaki Prefecture) as the northernmost geographical distribution in the Pacific; Matsuyama (Ehime Pref.); Shirahama (Wakayama Pref.); Amamioshima Island (Kagoshima Pref.); Tomari in Okinawa Island and Aka in Akajima Island (Okinawa Pref.). These are the first record of this species from each locality and the first record of this species in Japan. The morphological characteristics of the medusae caught in the sea generally agree with those reported earlier from other regions in the world. However, the medusae reared in the laboratory exhibited considerable morphological differences, which are highlighted here. Geographical distribution is also mentioned.

Introduction

The hydrozoan family Cirrholoveniidae Bouillon, 1984 contains two genera, *Cirrholovenia* Kramp, 1959 and *Paralovenia* Bouillon, 1984 (Bouillon, 1984a, b; 1985). Recently many medusae belonging to the former genus were collected by the author from six localities in Japan. They were identified as *C. tetranema* Kramp, 1959, being apparently different from *C. polynema* Kramp, 1959, the type species of the genus.

Some of these medusae were reared under controlled laboratory conditions until they became inactive or were attacked by the protozoans, and the morphological changes were observed. The cnidome, the gametes and spawning time of this species were observed for the first time. The morphological differences between the present specimens and those described from other regions of the world are pointed out.

Material and Methods

A total of 33 medusae were collected from six localities around Japan as described below (Fig. 1). Their morphology was observed immediately or within four days after capture under a stereoscopic and a light microscope. Some of them were brought back to the laboratory and reared in filtered seawater at 22°C in 60 ml polystyrene vessels under alternating 12h periods of light and dark until they became inactive. They were fed with *Artemia* nauplii. The seawater was changed every day.

From Tsushima Island, Nagasaki Prefecture, at the entrance of the Sea of Japan, two medusae were collected alive, scooping gently in a 60 ml polystyrene vessel by hand on August 20, 1992. They were swimming in a medusan swarm at subsurface depth along the coast of Takeshiki in Asou Bay (Fig. 1, T). The smaller specimen was preserved soon after collection and examined later (measurements were

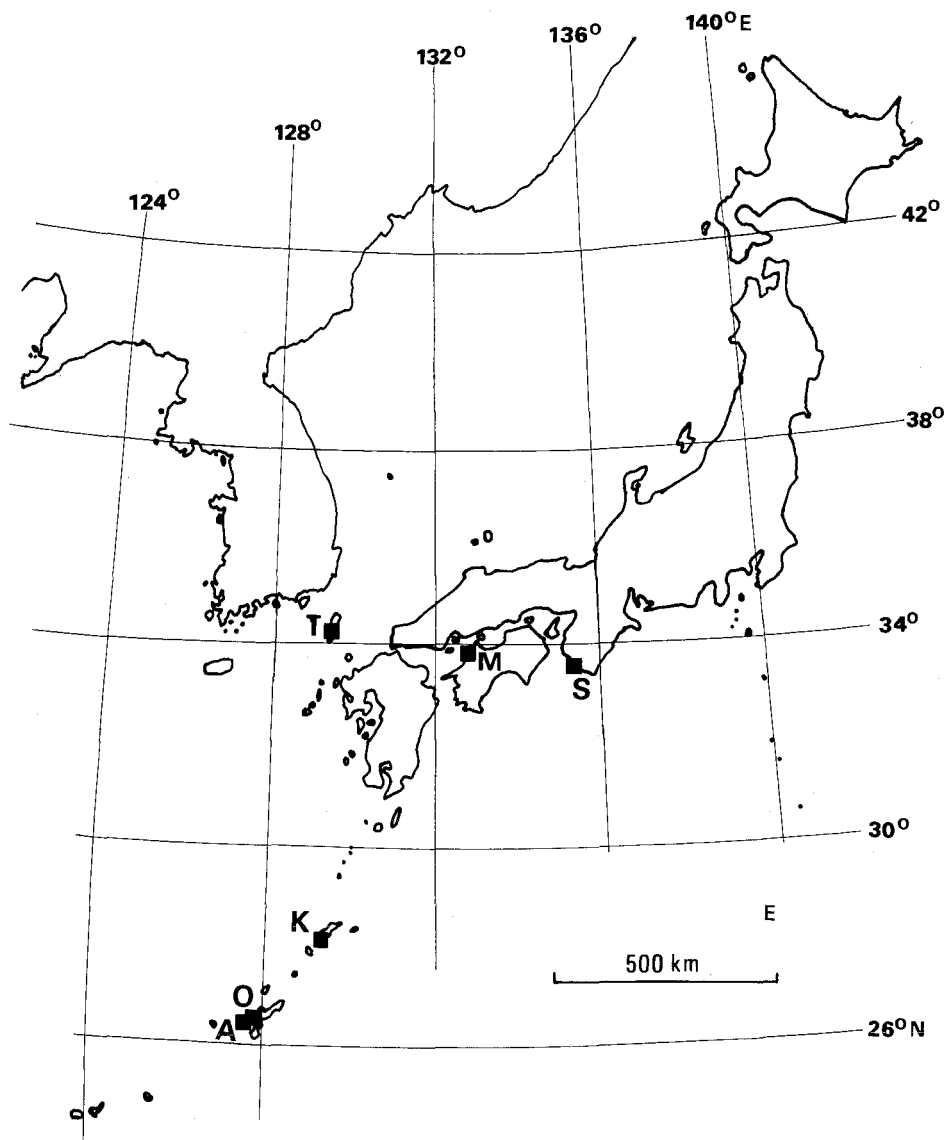


Fig. 1. Localities from which medusae of *Cirrholovenia tetranema* were collected. A: Akajima Island. K: Koniya, Amamioshima Island. M: Mitsuhamma, Ehime Prefecture. O: Tomari, Okinawa Island. S: Shirahama, Wakayama Prefecture. T: Takeshiki, Tsushima Island.

partly possible), while the larger one was brought alive to the laboratory. On the 4th day after collection it was anesthetized by $MgCl_2$ solution and examined in detail. On September 28, 1994 another specimen was collected from the same locality by towing a plankton net in the upper 8 m.

On August 7, 1993 three medusae were collected at Mitsuhamma port, Matsuyama, Ehime Prefecture in the Inland Sea of Japan by vertical towing of a plankton net at 10–0 m (Fig. 1, M). Their morphology was examined on the next day in the laboratory, and the growth of two medusae were observed together with the cnidome.

Two medusae were collected at Koniya port, Setouchi-cho, Amamioshima Island, Kagoshima Prefecture

by horizontal and vertical (from 8 m depth to surface) towing of a plankton net on October 11, 1993 (Fig. 1, K). The morphology of these specimens, one preserved *in situ* and the other reared in the laboratory, was observed.

At Shirahama, Wakayama Prefecture (Fig. 1, S: 33°41.5'N, 135°18.4'E) one specimen was collected by vertical (in the upper 30 m) towing a plankton net on December 2, 1993. It was examined immediately after collection.

At Aka port, Akajima Island, Okinawa Prefecture a total of 23 specimens were collected at 5–0 m depth during 21st to 23rd November, 1993 and 10th to 12th March, 1994 (Fig. 1, A). They were examined immediately or within a few days after collection. Some of these specimens were reared in the laboratory for a maximum period of one month to observe their gametes and also to record their spawning time.

One specimen caught in the upper 7 m depth on March 12, 1994 at Tomari port, Okinawa Island, Okinawa Prefecture was examined after two and seven days after its collection (Fig. 1, O).

The figures 2 and 6–8 were drawn with the aid of a drawing tube attached to a phase-contrast microscope. Photographs were taken in a living specimen (Figs. 3, 4).

Description

The morphology of a medusa caught in the sea is described first, following the synonymy. Then the growth of medusa is described together with the cnidome, the gametes, and spawning time as well as the remarks and the geographical distribution.

Cirrhovenia tetranema Kramp, 1959

(Figs. 2–8)

Cirrhovenia tetranema Kramp, 1959, pp. 253–254, Figs. 17a, b; —, 1961, p. 173; —, 1968, p. 80, Fig. 214; Brinckmann, 1965, pp. 13–15, Fig. 1; Brinckmann-Voss, 1987, p. 137; Allwein, 1967, p. 122, p. 129; Moreira, 1967, pp. 426–427; —, 1975, pp. 556–557; —, 1978, pp. 47–48; Navas, 1971, p. 5, p. 11, p. 17; —, 1981, p. 235, pp. 254–255; Goy, 1972, pp. 991–992; —, 1979, p. 276, Fig. 14; —, Lakkis & Zeidane, 1990, p. 82, 86; —, — & —, 1991, pp. 113–114, 123–124, Fig. 35; Bouillon, 1978, p. 125; —, 1984b, p. 27; Xu & Zhang, 1981, p. 374, p. 377, p. 382, Fig. 8; Mao & Jinbiao, 1991, p. 304; Lakkis, 1991, p. 125.

Eugymnanthea minuta Uchida, 1964, pp. 101–104, pp. 106–107, Figs. 1a, 1b, Figs. 2, 3; Rees, 1967, p. 221.

Morphology of medusa caught in the sea

The umbrella is up to 1.35 mm in diameter and 1.25 mm in height (Table 1). The mesogloea is generally thin, with a maximum thickness of 0.13 mm at the umbrellar apex. In an immature medusa exumbrellar nematocysts are sparsely found (Fig. 2, A). The manubrium is tubular and short, up to 0.35 mm long. No peduncle is present. Four oral lips are simple. Four tentacles extend from conical tentacular bulbs. No marginal warts are present (Figs. 2, 3). There are usually four, rarely five or six statocysts, and one statocyst in each of the interradiar portion of the umbrellar margin, but sometimes near the tentacular bulbs (Fig. 2, A). Each statocyst contains a single statolith but very rarely two are also present (Fig. 2, A). Lateral cirri are absent. In each quadrant a maximum number of seven marginal cirri were found. In a medusa up to 26 cirri were observed. Four gonads extend from the stomach base along the four radial canals, but do not reach the umbrellar margin. The gonads are smooth. The medusa is transparent.

Table 1. Some taxonomic characters of the medusa of *Cirrhlovenia tetranema* caught in the sea around Japan. Measurements were taken within four days after collection. Blank: Not measured due to modification of shape.

Sex	Diameter (mm)	Height (mm)	Thickness of jelly at umbrellar apex (mm)	Length of manubrium (mm)	Total number of stato- cysts	Total number of stato- liths	No. of marginal cirri in each quadrant (or in two bulbs)
Takeshiki, Tsushima Is.							
Female	0.93	0.63	0.08	0.13	4	4	5, 4, 4, 4
?	0.75	0.48	0.03	0.10	4		3, 3, 3, 3
?					4	4	max. 7
Mitsuhama							
Male	0.88	0.65	0.08	0.25	4	4	5, 2, 4, 3
?	0.88	0.63	0.13	0.15	4	5	3, 2, 1, 2
?	0.78	0.50	0.08	0.15	4	4	2, 2, 2, 2
Shirahama							
?	1.20	0.98	0.05	0.35**	6	6	4, 4, 4, 3
Koniya, Amamioshima Is.							
Male	1.30	1.25	0.05	0.23	4	3	5, 5, 4, 4
Male					5	5	6, 4, 4, 4
Tomari, Okinawa Is.							
?	0.83				3	2	3, 3, 2, 2
Aka, Akajima Is.							
Female	1.35				4	4	4, 4, 3, 3
Female	1.08	0.90	0.08		4	4	3, 3, 2, 2
Female	1.02	0.78	0.05	0.24	4	4	6, 5, 2, 3
Female					4	4	7, 6, 7, 6
Female*					3	3	6, 6, 6, 5, 2
Male	1.35	1.00	0.10		4	4	5, 4, 4, 4
Male	1.23	0.88			4	4	4, 4, 4, 3
Male	1.12	0.80	0.07	0.24	4	4	6, 3, 5, 5
?	1.30	0.98	0.05		4	4	4, 3, 3, 3
?	1.28				4	4	4, 4, 3, 3
?	1.28	0.85			4	4	3, 3, 3, 2
?	1.10	0.93	0.05		4	4	5, 4, 4, 4
?	1.10	0.88	0.10		4	4	4, 4, 4, 3
?	1.05	0.80	0.08		4	4	3, 2, 3, 2
?	1.03	0.93	0.05		4	4	4, 4, 3, 3
?	0.98	0.80	0.07	0.24	4	4	5, 4, 4, 4
?	0.98	0.78	0.05	0.20	4	3	3, 3, 3, 3
?	0.90	0.63	0.07	0.15	4	4	4, 4, 4, 4
?	0.90	0.83	0.07	0.22	4	4	4, 4, 4, 3
?	0.90	0.73			4	4	3, 3, 3, 2
?	0.90	0.75	0.08		4	4	3, 2, 2, 2
?	0.88	0.68	0.05		4	4	3, 3, 2, 2
?*	1.05	0.78	0.08		5	5	4, 3, 3, 3, 3

*: Specimens with five radial canals. **: Larger one of the two manubria.

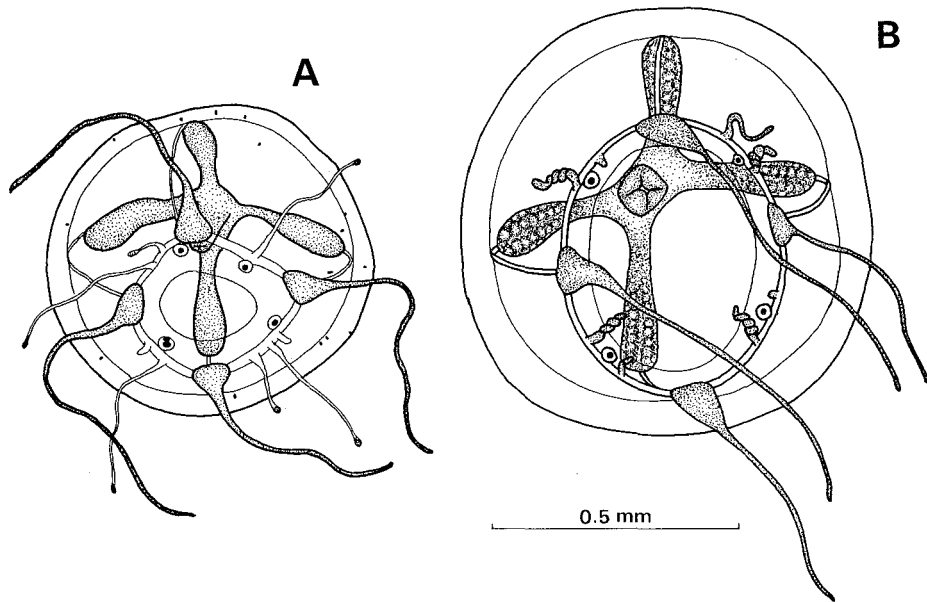


Fig. 2. Morphology of the medusa of *Cirrholovenia tetranema*. A: Oblique view of immature specimen from Mitsuhamma. Note the position of two statocysts which are not found interradially, one statocyst containing two statoliths, and the presence of the exumbrellar nematocysts. B: Oral view of a female specimen from Tsushima Island. Note that decrease in number of the marginal cirri within four days after collection (see Table 1).

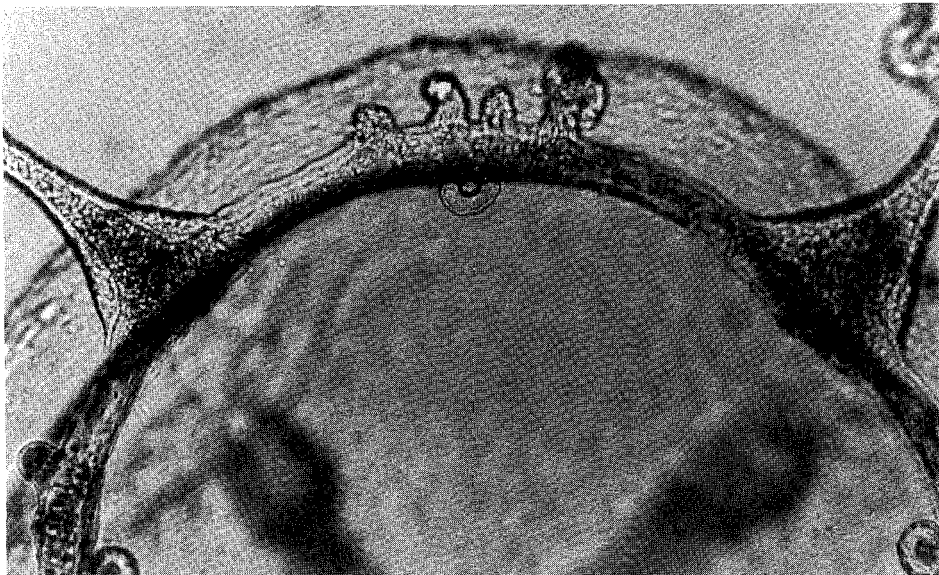


Fig. 3. Part of umbrellar margin of *Cirrholovenia tetranema* from Akajima Island (Aboral view).

As an exception, one specimen collected from Shirahama had two manubria and two specimens collected from Akajima Island had five radial canals (Table 1). Although the female specimen from Akajima Island had five radial canals and five tentacular bulbs, it had only three statocysts as the minimal number of statocysts per medusa.

Growth of medusa in the laboratory

Medusae caught in the sea usually grow in the laboratory even if after maturation. A medusa collected from Mitsuhamma grew up to 1.28 mm in diameter after 10 days of its collection (Fig. 5, B). Another one from Amamioshima Island (Fig. 4) grew up to 2.38 mm in diameter after 13 days (Fig. 5, C) and a female specimen from Akajima Island grew to 2.50 mm in diameter after 22 days.

The marginal cirri increased in number in every specimen (Fig. 5, A-C), reaching to a maximum of 51 per medusa (Fig. 5, C). The maximum number of marginal cirri in a quadrant was 15 (Tables 1, 2).

The number of tentacles became five due to the formation of one tentacular bulb interradially in a medusa from Amamioshima Island (Table 2).

The number of statoliths per statocyst is usually constant (1 per statocyst), while the number of statocysts increased, reaching to a maximum of nine (Fig. 5, C). The maximum number of statoliths per statocyst was four in a 33-day-old female specimen from Akajima Island, which grew as the largest specimen in the present study (2.50 mm in diameter). A total of 17 statoliths were found in this specimen (each statocyst contained 3, 2, 2, 2, 2, 4, 1, 1 statoliths).

The four oral lips remains simple (Fig. 6, B).

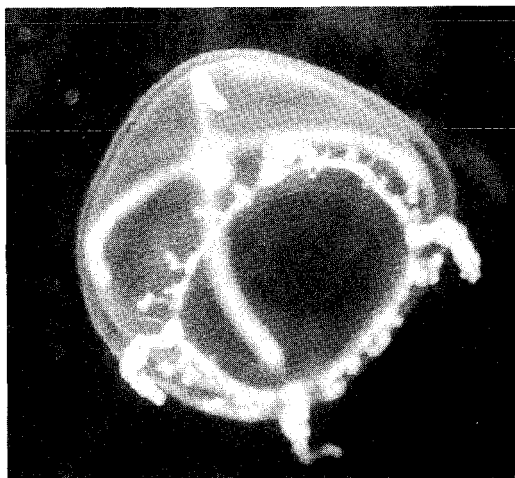


Fig. 4. A spent male medusa of *Cirrholovenia tetranema* reared for eight days after collection at Amamioshima Island (2.08 mm in diameter).

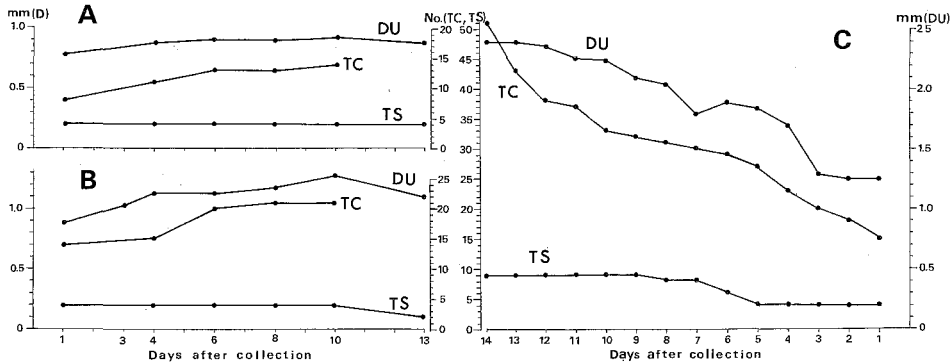


Fig. 5. Growth of three medusae of *Cirrholovenia tetranema*, two from Mitsuhamma (A, B) and one from Amamioshima Island (C). DU: Diameter of umbrella. TC: Total number of the marginal cirri per medusa. TS: Total number of the statocysts per medusa.

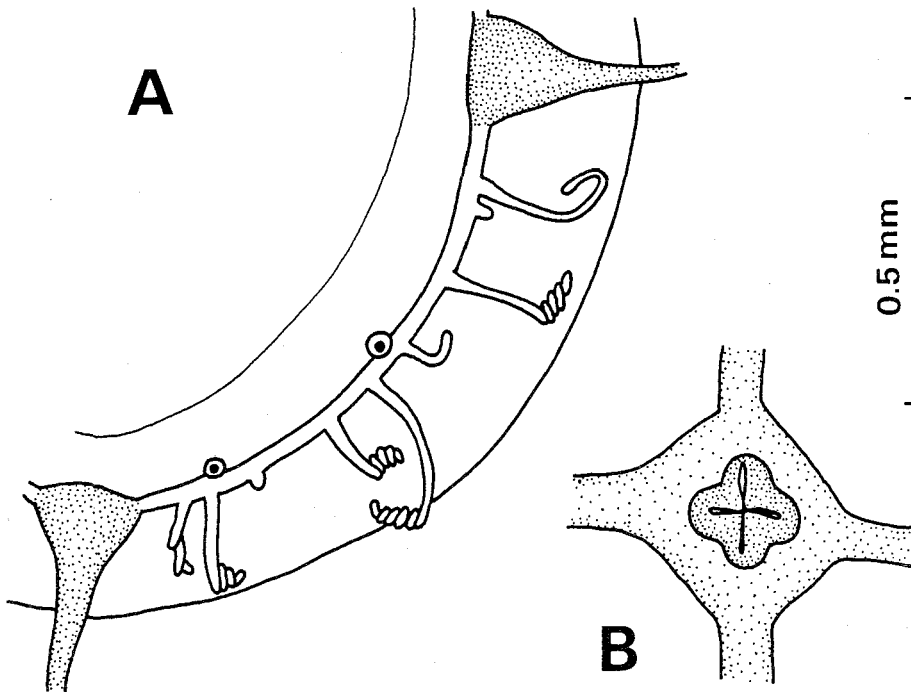


Fig. 6. Body portions of a laboratory-reared medusa of *Cirrholovenia tetranema* from Amamioshima Island. A: Umbrellar margin of a quadrant, showing two statocysts and nine marginal cirri (7-day-old stage). B: Crusiform oral lips and the broad stomach (14-day-old stage).

Regeneration

Regeneration of the tentacular bulbs, statocysts and marginal cirri was observed in one male specimen from Akajima Island on the 4th day after nearly half of body

Table 2. Daily change of the number of marginal cirri, statocysts, and of the tentacular bulbs in each quadrant of one medusa from Amamioshima Island (see Fig. 5, C).

Age (days)	Number in each quadrant:		
	marginal cirri	statocysts	tentacular bulbs
1	3, 4, 4, 4	1, 1, 1, 1	1, 1, 1, 1
2	4, 4, 5, 5	1, 1, 1, 1	1, 1, 1, 1
3	5, 5, 5, 5	1, 1, 1, 1	1, 1, 1, 1
4	5, 6, 6, 6	1, 1, 1, 1	1, 1, 1, 1
5	6, 7, 7, 7	1, 1, 1, 1	1, 1, 1, 1
6	6, 8, 7, 8	1, 1, 2, 2	1, 1, 1, 1
7	6, 7, 8, 9	2, 2, 2, 2	1, 1, 1, 1
8	7, 8, 7, 9	2, 2, 2, 2	1, 1, 1, 1
9	8, 8, 7, 9	2, 2, 2, 3	1, 1, 1, 1
10	8, 8, 8, 9	2, 2, 2, 3	1, 1, 1, 1
11	8,10, 9,10	2, 2, 2, 3	1, 1, 1, 1*
12	9,10,10, 9	2, 2, 2, 3	1, 1, 1, 1*
13	10,12,10,11	2, 2, 2, 3	1, 1, 1, 2
14	15,14,10,12	2, 2, 2, 3	1, 1, 1, 2
15	9,12,10, 5	2, 2, 2, 3	1, 1, 1, 2
16			

*: One marginal wart was produced besides four tentacular bulbs.

-: Data unavailable due to degeneration by attack of protozoans.

was cut off for observation of the sperm (see below). This specimen with its intact manubrium fed on *Artemia* larvae and produced a total number of six tentacles, seven statocysts, and ten statoliths (three statocysts contained two statoliths) on the 9th day of its regeneration. Thus it appeared as a medusa of different morphology from the initial ordinary one.

Cnidome

Two kinds of nematocysts are present (Fig. 7, Table 3), though the exumbrellar nematocysts were not examined. The largest nematocysts are on the terminal knob of the marginal cirri; they belong to the basitrichous isorhizas (Fig. 7, Bb). Their number in each cirrus was 11–19 with the mean and standard deviation of 15.8 ± 2.1 ($N=14$, examined in two medusae). The smallest nematocysts are on the distal portion of the marginal cirrus but not on its terminal knob; they belong to the atrichous isorhizas (Fig. 7, A). Two types of atrichous isorhizas, a larger type and a smaller one (Fig. 7, Cc, Dd), are present on the tentacles. The basitrichous isorhizas are also present on both tentacle and manubrium (Fig. 7, Ee, F).

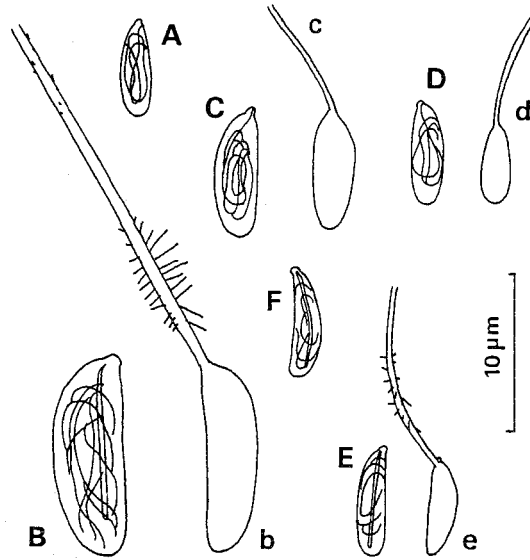


Fig. 7. Nematocysts of the medusa of *Cirrholovenia tetranema*. A, Cc, Dd: atrichous isorhizas; Bb, Ee, F: basitrichous isorhizas. A, Bb: on the marginal cirri; Cc-Ee: on the tentacles; F: on the manubrium.

Table 3. Distribution of nematocysts of medusa of *Cirrholovenia tetranema*. Dimensions are length x maximum width of undischarged capsules, in μm . Dimensions are mean \pm SD, range and the number of nematocysts and medusae examined.

	Atrichous isorhizas	Basitrichous isorhizas
Tentacles	$8.1 \pm 0.3 \times 2.9 \pm 0.1$, 21, 2 (7.6–8.8) (2.8–3.0) $6.6 \pm 0.6 \times 2.3 \pm 0.2$, 25, 2 (5.8–7.6) (2.0–2.6)	$7.0 \pm 0.2 \times 2.0 \pm 0.1$, 27, 2 (6.4–7.6) (1.8–2.2)
Marginal cirri	$4.8 \pm 0.7 \times 1.8 \pm 0.2$, 22, 2 (4.0–6.4) (1.6–2.0)	$13.3 \pm 0.6 \times 4.5 \pm 0.2$, 26, 3 (12.4–14.6) (4.4–4.8)
Manubrium	Absent	$7.0 \pm 0.2 \times 2.0 \pm 0.1$, 15, 1 (6.4–7.2) (1.8–2.0)

Gametes and spawning time

The size of sperm was measured in one male medusa from Akajima Island on a few days after collection. The length of head and middle piece together was 2.6–2.8 μm (N=4) and the greatest width was 1.8–2.0 μm (N=4). The tail was 46–56 μm in length (N=5).

The diameter of unfertilized eggs was measured on five occasions immediately

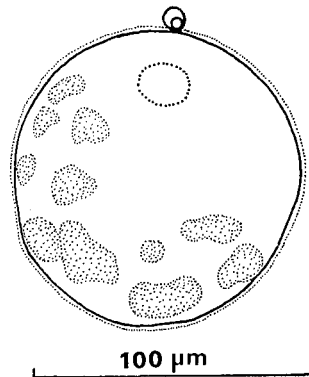


Fig. 8. Unfertilized egg of *Cirrholovenia tetranema*, immediately after spawning. Note many opaque inclusions.

after discharge from the gonads of a female medusa from Akajima Island. The diameter varied from 97.6 to 112.0 μm with the mean and standard deviation of $103.5 \pm 3.9 \mu\text{m}$ ($N=25$). Spawning occurred approximately 60 minutes after dark. The eggs spawned were not entirely transparent, but contained many opaque inclusions (Fig. 8).

Remarks

The synonymy of *Eugymnanthea minuta* Uchida from the Gulf of Naples with the present species was already mentioned by Kubota (1978, p. 141). Uchida's (1964) two specimens are the first record of *Cirrholovenia tetranema* in the Mediterranean and the second record of this species in the world.

The number of statolith per statocyst of the present specimens caught in the sea is usually less than that of the above-mentioned specimens from the Mediterranean described by Uchida (1964), while the same as that in the specimens collected from Brazil by Moreira (per. comm.) (see Table 4). However, in some statocysts of the laboratory-reared specimens the number increased up to four, overlapping with the number described in the aforementioned specimens from the Mediterranean.

As the number of statocysts is reported to be variable (up to eight) in the original description (Kramp, 1959), the number was demonstrated as a variable character in the present specimens (Table 2). However, most of the specimens grown in the sea (28 out of 31) had four statocysts irrespective of the size of medusa and sex. A specimen from Tomari that had three statocysts on capture (Table 1) produced a new statocyst on the 7th day, and thus had the normal number of statocysts.

The gonads in the present specimens extend from the base of the stomach as in the holotype from the Solomon Islands (Kramp, 1959) and those from Southeast Asia (Kramp, 1959), Mediterranean (Uchida, 1964; Brinckmann, 1965; Goy, 1972; Goy, Lakkis & Zeidane, 1991), North American Atlantic (Allwein, 1967), and the South American Atlantic (Moreira, 1967, 1975). However, some specimens collected from the Mediterranean (Uchida, 1964), the Brazilian coast (Goy, 1979), the Chinese

Table 4. Morphological comparison of the medusa of *Cirrhovolenia tetranema* around the world.

Locality (see Fig. 9)	Umbrellar diameter (mm)	No. of stato- cysts	No. of stato- liths/ statocyst	No. of marginal cirri/ quadrant	Connection of gonads with the stomach	No. of medusae examined	References
Solomon Islands* & SE Asia	below 1.5	4* or 8	-	up to 8*	connected*	7 ²⁾	Kramp, 1959
Papua New Guinea	below 1.4	4, rarely 7	x	up to 9	connected or separated	50 ^{2,3)}	Bouillon, 1984b
Italy	0.88–0.99	4 or 6	3–4	4–9	connected or separated	2 ³⁾	Uchida, 1964
Italy	1.7**	4	-	4–5**	connected**	1 ³⁾	Brinckmann, 1965
France	below 1.0	4, rarely 8	-	up to 6 ¹⁾	connected	15	Goy, 1972
Lebanon	2**	4 or more	-	7	connected	20	Goy, Lakkis & Zeidane, 1991
USA	2.8	4	-	6–8	connected	1	Allwein, 1967
Brazil	below 1.7	4	1	up to 8	connected	40	Moreira, 1975, per. comm.
Brazil	below 1.5	4	-	8**	separated	2 ²⁾	Goy, 1979
China	at least 3.2**	4	-	7–8	separated	7	Xu & Zhang, 1981
Japan	below 2.50	usually 4, up to 9	usually 1, up to 4	up to 15	connected	33 ^{2,3)}	Kubota, present study

*: Type locality and the character state of the holotype.

**: Read from the figure of the paper.

-: No description. x: unavailable.

1): 24/4.

2): At least one specimen is female.

3): At least one specimen is male.

coast (Xu & Zhang, 1981), and from the Papua New Guinea (*cf.* Bouillon, 1984b) have gonads separated from the stomach. Such a difference in the position of the gonads, irrespective of the size of medusa (see Table 4), can be linked with the physiological status of medusae. As a matter of fact, it was observed in the present

medusae reared in the laboratory that the proximal portion of radial canal became swollen on swallowing the food, and as a result of such swellings the gonads appeared as if they were those directly connected with the stomach.

The marginal cirri are very variable in number irrespective of the size of the mature medusa, and up to 15 cirri have been found in a quadrant (Tables 2, 4). According to physiological conditions the number easily changes as was demonstrated in the present rearing.

As in the case of Mediterranean specimens (Brinckmann, 1965), the present medusa is supposed to have shed eggs in the early morning since it spawned eggs around one hour after dark. But, Brinckmann (1965) observed the other spawning of eggs during the night.

The primary hydroid and a very tiny, stolonial colony were obtained from the medusae with the gonads extending from the stomach base by the laboratory-rearing by Brinckmann (1965, pp. 13–14, Figs. 2, 3) and also by Moreira (1975, p. 557). Their hydroids of *C. tetranema* surely belong to the Thecata and are placed in the hydroid genus *Cuspidella* Hincks, 1868 by Brinckmann (1965). Therefore, Uchida's (1964) conjecture that the hydroid of *C. tetranema* (originally described as *Eugymnanthea minuta*) is misleading. It was observed that from the hydroids commensal with *Mytilus edulis galloprovincialis* in Italy medusae of *Eugymnanthea inquilina* Palombi are exclusively liberated (Kubota, 1989; Piraino et. al., 1994).

Geographical distribution

The present new record in Asou Bay, Tsushima Island is the northernmost

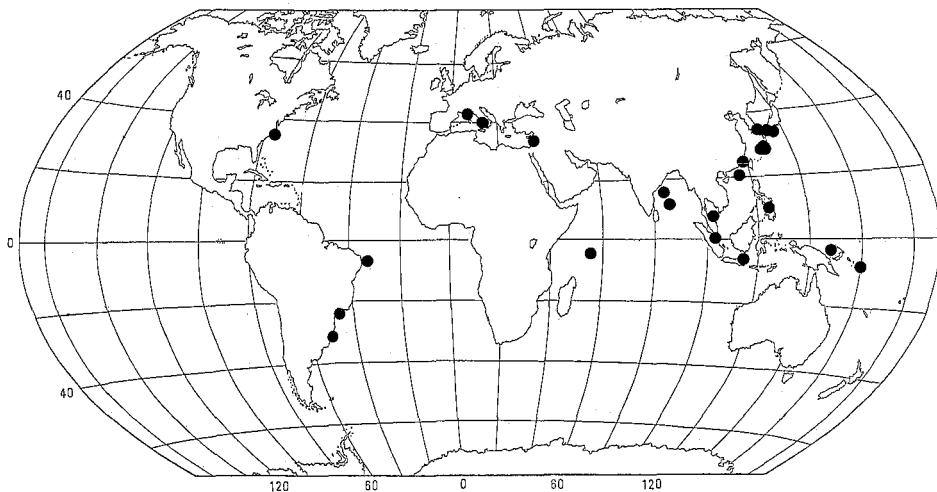


Fig. 9. Geographical distribution of the medusa of *Cirrholovenia tetranema*, compiled from Kramp (1959), Uchida (1964), Brinckmann (1965), Allwein (1967), Navas (1971), Moreira (1975), Bouillon (1978, 1984b), Goy (1972, 1979), Navas-Pereira (1981), Xu and Zhang (1981), Brinckmann-Voss (1987), Goy, Lakkis & Zeidane (1991), Mao & Jinbiao (1991), and the present study.

geographical distribution of this species in the Pacific (Fig. 9). All other specimens in Japan are also new localities (see Fig. 1). Furthermore, the present species is a new member in the Japanese fauna.

The previous records of the medusae of *Cirrhlovenia tetranema* are the following (Fig. 9): near Solomon Islands (Kramp, 1959), the South China Sea (Kramp, 1959; Xu & Zhang, 1981); the Taiwan Strait (Mao & Jinbiao, 1991); the Gulf of Siam (Kramp, 1959); the Java Sea (Kramp, 1959), the Strait of Malacca (Kramp, 1959); the Bismark Sea, Papua New Guinea (Bouillon, 1984b); the Indian Ocean (Navas, 1971; Bouillon, 1978); the North American Atlantic (Allwein, 1967); the South American Atlantic (Moreira, 1967, 1975; Goy, 1979; Navas, 1981); the Mediterranean (Uchida, 1964; Brinckmann, 1965; Goy, 1972; Brinckmann-Voss, 1987; Lakkis, 1991; Goy, Lakkis & Zeidane, 1991). Therefore, *C. tetranema* appears to have a wide distribution extending from the tropics to the temperate regions. It is noteworthy that the medusae have not been recorded from the east coasts of the Pacific and the Atlantic Ocean.

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